



STUDYING THE EFFICIENCY OF *CANNA GENERALIS* AFTER BEING INOCULATED WITH MYCORRHIZA FUNGUS (*GLOMUS MOSSEA*) IN PLANT TREATMENTS CONTAMINATED WITH LEAD AND CADMIUM

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Abstract

The experiment was conducted in greenhouse of Department of Horticulture and Landscaper Gardening, College of Agriculture, University of Baghdad in Al-Jadiriya for the spring season 2016-2017. The soil was contaminated with concentrations of lead element (0, 75, 150 mg.kg⁻¹) and concentrations of Cadmium (0, 10, 20 mg.kg⁻¹). The results showed that the contamination of the soil with lead and cadmium resulted in a decrease in the nutrient elements (Nitrogen, Phosphorus and Potassium), while the nutrient elements were increased when plant root was inoculated with Mycorrhiza fungi. The results showed that the accumulation of heavy metals was increased by increasing the added concentrations. It was noticed that the highest accumulation of lead and cadmium in root group was (74 mg.kg⁻¹) at the concentration of (150 mg.kg⁻¹, the 03.11 mg.kg⁻¹) lead element at a concentration of (20 mg.kg⁻¹) cadmium. In addition, lead and cadmium in the soil increased after the end of the experiment by increasing the added concentrations.

Key words : *Canna generalis* plant, mycorrhiza fungus, lead, cadmium.

Introduction

Nature features is changed in many parts of the world, especially with the emergence of the industrial revolution, modern technologies and population expansion, without taking into account their impacts on the environment. This has resulted in negative impacts on environmental systems and the most dangerous is environmental pollution, which is progressing with scientific progress wherever it goes. It entered with the human into its a dwelling place and arrived at his food mixed with water, air and soil, which raised concern and made the issue of environmental pollution and tampering with the natural environment of the important issues at present time. It was a serious endeavor to repair the polluted environments. Among the pollutants of environmental importance for its serious adverse effects, in addition to its numerous and wide industrial use are the heavy elements. The most important of these elements are the spread of lead and cadmium. These two elements are the predominant elements of the various sources of

industrial activity such as mining, Metal melting, automotive exhausts, dyes and agricultural applications such as fertilizers, pesticides, sewage and others (Ali *et al.*, 2012). Phytoremediation is a form of biologic treatment where plant species with the ability to absorb and accumulate relatively large quantities of these pollutants are selected by reservation, removing or analyzing different contaminants in their tissues (Ejaz *et al.*, 2007). Plant treatment technology can be categorized on the basis of treatment mechanisms: the removal of pollutants from the soil and their concentration in plant tissues or the cracking of polluting elements by various biological and non-biological processes in plants such as volatilization or restriction and paralyzing of pollutant elements in the root zone (EPA, 2000). Plant processing technology is one of the important environmental techniques by which pollutants can be removed from the environment. The plant species that have the ability to absorb and accumulate large quantities of these pollutants are selected without affecting their growth. Therefore, these plants are used for other purposes such as fencing

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around industrial areas or landscaping gardening and fields. They can also be used to plant the streets and roads to get rid of the largest amount of pollutants in the environment (Aba Al-khayl *et al.*, 2013). Among these plants using in the treatment are *Canna generalis*, which belongs to the Cannaceae family, which there are about 50 plant species that flowering most of the year except winter. It is a summer perennial plant, native to central America, where these plants have the ability to absorb and accumulate heavy elements in its various parts of plant (Al-batal, 2010). Mycorrhiza is believed to have an added role in increasing the accumulation of heavy elements in the plant. This agrees with Al-Atabi and Mahdi, 2015). It was found that the extrudical hyphae of the Mycorrhiza is responsible on the transfer of Cadmium from the soil to plant when inoculation by Mycorrhiza fungi, Mycorrhiza fungi play a significant role in the removal of contaminants from soils subjecting to contamination with heavy metals or organic pollutants. The effect of Mycorrhiza varies depending on the concentration and type of pollutant (Cobbett and Goldsbrough, 2000). The study aims to test the efficiency of the generala *Canna* plant to treat the soils contaminated with heavy elements (lead and Cadmium), to determine which of plant parts (shoot group or root group) accumulation of the highest percentage of these heavy elements and know the role of Mycorrhiza fungi in reducing the toxicity of these elements.

Materials and Methods

The experiment was conducted in greenhouse of Department of Horticulture and Landscaper Gardening, College of Agriculture, University of Baghdad in Al-Jadiriya for the spring season 2016-2017. Black polyethylene bags were utilized sterile superficially with a solution of sodium hypochlorite, with a concentration of 2% for 10 kg capacity of soil, a layer of gravel were placed in the bags with a height of 4 cm. The soil was brought from one of the banks of the Tigris River in Al-Jadiriya region. The soil was mixed with a quantity of peat moss, with a ratio of 3: 1. The 7 kg of soil were weighed for each bags preparing for cultivation and contaminated with the salts of contaminated elements (lead acetate and Cadmium chloride) and with three concentrations per element. A factorial experiment was conducted within Randomized Complete Block design, with three factors:- The first factor is soil contaminate with lead element, with three levels (0.75, 150 mg.Kg⁻¹), which is symbolized by (Pb₀, Pb₁, Pb₂), the second factor is soil contaminate with Cadmium, with three levels (0, 10, 20 mg.kg⁻¹), which is symbolized by (Cd₀, Cd₁, Cd₂),

the third factor is Mycorrhiza fungi (without inoculating plants and inoculated plants), with two levels of the addition of Mycorrhiza fungi to root of the plant, which is symbolized by (M₀, M₁), respectively and with three replicates. The averages of the treatments were compared with the least significant difference (LSD) at a significant level of 0.05 and after 14 days of soil contamination process with salts of heavy elements (Khamis *et al.*, 2014). The bags were cultivated with seedlings of *Canna generalis* plant with 8-10 cm long and 2-3 leaves on 20 March 2017. The seedlings were divided into two parts: first part inoculated by the fungal isolation (spores, mycelia, infected roots and soil), 15 g of each bag in form of pad, So that it touches the roots of the plant and according to the method described by Al-Yahya'ei *et al.*, 2011) at a depth of about 10 cm. The second part was not inoculated; the plants were irrigated to the limits of the field capacity. Irrigation was re-applied when the soil depleted 25% of the prepared water and according to the weight method because the plant is preferred high moisture until the experiment ends 20/2/2018.

Study indicators

Nitrogen N% : Nitrogen is estimated by Kejl Dahl-Micro (Jones and Steyn, 1973).

Phosphorus element P% : Phosphorus was estimated using ammonium molybdate and vitamin C and Absorption was measured at 620 nm and according to the method used by Schutzendubel and Polle, 2002).

Potassium element K% : Potassium is estimated by Flamephotometer (Yobouet *et al.*, 2010).

Measuring the concentration of lead and Cadmium elements in the plant for both shoot group and root group

Plant samples were washed, shoot group was separated from roots system. The samples were dried in an electric oven at 70°C and until the weight was fixed, then grinded with an electric mill. Each sample was weighed 0.2 g, placed in a 25 mL flask and then added 3 ml of concentrated sulfuric acid and 2 ml of pyrochloric acid. Samples were left for 24 hours and samples were then heated until colorless extracts were obtained. Samples were placed in a 50 mL volumetric flask, supplemented with distilled water and prepared for measurement by the Atomic Absorption Spectrophotometer (AAS) (Al-Sahaf and Fadel, 1989).

Measurement of heavy elements in soil after cultivating

The experiment plants were collected after 6 months

of cultivating. Soil samples were taken from each bag after cultivating. The concentrations of total heavy metals (lead and Cadmium) were estimated according to the method adopted by the US Environmental Protection Agency (EPA) in 1986 that mentioned in soil and plant analysis book [Laboratory index (Issam and Sayegh, 2007)].

Results and Discussion

Shoot group content of the nitrogen element (N%)

Table 1 show that the treatment of plants with lead element has a significant effect on the percentage of nitrogen. It was observed that with the increase of lead element concentrations, the percentage of nitrogen in shoot group decreased to 1.49% when it was treated with concentration of (150 mg.kg⁻¹) compared to Pb₀ which recorded 1.90%. The Cadmium treatments were also affected by the percentage of nitrogen, where the control treatment Cd₀ was excelled by giving it a value 1.93% compared to the Cd₂ treatment, which recorded 1.35%. The heavy elements may be linked with functional groups in the enzymes by stable bonds and formation of complexes, then disrupt the molecules that direct photosynthetic reactions or the heavy elements may concentrate on the cell membrane, Which changes its structural composition, It causing the impedes the exchange of ions and organic substances such as proteins and sugars or prevents them entirely from transition (Abd Al-Moneim *et al.*, 2012). Plant inoculation was accompanied with Mycorrhiza to obtain a significant increase in the percentage of nitrogen. The inoculation treatment M₁ gave the lowest percentage of nitrogen was 1.77%. While non-inoculation treatment M₀ recorded the lowest percentage of nitrogen was 1.55%. The increase in nitrogen in inoculated plants may be due to the role of Mycorrhiza fungi in increasing the absorption surface and thus increasing the absorption of water and nutrient elements as Mycorrhiza fungi have an effect in the process of fixing the bacteria for nitrogen and processing them with phosphorous necessary to meet their energy needs (Ishac, 2000). Pb₀Cd₀ and Pb₀Cd₁ were significantly excelled on the other interaction treatments in the lead-Cadmium concentration (2.19, 2.18%), respectively, while Pb₂Cd₂ gave the lowest nitrogen value of 1.27%. As for bi-interaction between lead element and Mycorrhiza, the treatment of M₁Pb₀ was significantly excelled on the other treatments by recording a percentage of 2.07% compared to M₀Pb₂ treatment, where the percentage of nitrogen decreased to 1.42%. The results of interaction between Cadmium and Mycorrhiza isolates showed that M₁Cd₀ was

significantly excelled by giving a percentage 2.07% on M₀Cd₂ treatment, which recorded the lowest value of 1.31%. The results of the triple interaction between lead, Cadmium and Mycorrhiza, M₁Pb₀Cd₁ and M₁Pb₀Cd₀ were significantly excelled by giving a value 2.47% and 2.36% compared to M₀Pb₂Cd₂, where the percentage of nitrogen decreased to 1.24%.

Shoot group content of phosphorus (P%)

Table 1 shows the results of the statistical analysis of the percentage of phosphorus element in shoot group. The concentration of lead were effected in the percentage of phosphorus, Pb₀ and Pb₁ treatments were significantly excelled by giving them the highest percentage of phosphorus (0.322%, 0.317%), respectively. While the increased concentration of lead in Pb₂ treatment led to reduce the percentage of phosphorus 0.286%. The reduction may be due to the replacement of heavy elements rather than a necessary element in the molecules responsible for photosynthesis (Sharma and Subhadra, 2010). In terms of Cadmium treatments, soil contamination with Cadmium concentrations led to excelling Cd₀ and Cd₁ treatments, which recorded (0.326, 0.317%) respectively compared to Cd₂, which gave the lowest content of phosphorus 0.281%. The reason for the low phosphorus concentration may be due to high concentrations of heavy metals may be due to the accumulation of heavy elements on the cell membrane, which may alter its composition and cause the exchange of ions or nutrients to be hindered and prevented from moving (Abd Al-Moneim *et al.*, 2012). Inoculation treatment by Mycorrhiza (M₁) was significantly affected in this trait, which giving 0.335% compared to non-inoculated treatment M₀, which recorded 0.281%. This is due to Mycorrhiza fungi that have an important role in increasing the phosphorus availability in the soil by reducing the pH and increasing the absorption of the element through the outer mycelia, thus increased the phosphorus component in the plants inoculated with Mycorrhiza fungi. Pb₁Cd₁ and Pb₀Cd₀ treatments were significantly excelled in the bi-interaction between lead and Cadmium element, recording 0.343 and 0.335% respectively. Also, it did not differ significantly from the Pb₀Cd₁ treatment, which recorded value of 0.330%, while the highest reduction in percentage of phosphorus was at Pb₂Cd₂ treatment which amounted to 0.248%. The bi-interaction between lead element and Mycorrhiza showed a significant excelling between M₁Pb₀ and M₁Pb₁ treatments on the rest of the treatments by giving it (0.352, 0.346%) while M₀Pb₂ gave the lowest percentage of phosphorus 0.263%. As for the effect of the bi-interaction between Cadmium and Mycorrhiza, M₁Cd₀ and M₁Cd₁

treatments were significantly excelled by giving them (0.358, 0.345%) while the percentage of phosphorus decreased to 0.260% at M_0Cd_2 treatment. In addition, the results of the triple interaction between the study factors showed that the $M_1Pb_0Cd_0$, $M_1Pb_1Cd_0$ and $M_1Pb_1Cd_1$ were significantly excelled on the rest of the treatments by recording a values of (0.370, 0.370, 0.366), respectively. The $M_1Pb_0Cd_1$ did not differ significantly from the above mentioned treatments, which gave value was 0.353 and the highest decrease in phosphorus was obtained at $M_0Pb_2Cd_2$ with 0.223%.

Shoot group content of Potassium (K%)

Table 1 shows the results of the statistical analysis of the potassium element. It was observed that the contamination of the plant with lead element resulted in a significant decrease in the percentage of potassium in shoot group potassium element was decreased in the treatment of Pb_2 to 2.85% compared to the control treatment Pb_0 which gave the highest percentage of potassium 3.34%. Potassium deficiency may be due to replacing the element of lead rather than potassium in the molecules (Sharma and Subhadra, 2010). The Cadmium treatments were significantly affected by this effect, as the potassium component decreased with the increase of used Cadmium concentrations. It was noted

that the highest decrease in the potassium element was recorded at the Cadmium concentration of 20 mg.Kg⁻¹ which reaching of 2.76% while the Cd_0 treatment gave the highest percentage of potassium 3.50%. The decrease in potassium may be due to the flow of potassium ions outside the cells of plasma membrane (Andon *et al.*, 2005) as a result of the addition of heavy elements and therefore the greater concentrations of heavy elements less absorption of nutrient elements, including the element of potassium by the plant. A significant effect was observed when plants were inoculated with Mycorrhiza. Mycorrhiza treatment M_1 was significantly excelled in increasing percentage of potassium by giving it a value of 3.77% compared to plants non-inoculated with Mycorrhiza M_{03} , which recorded 2.43%. In bi-interaction between the study factors (lead and cadmium), Pb_0Cd_0 and Pb_1Cd_0 treatments were significantly excelled in the increasing potassium element in shoot group of plant by recording them a value (3.72, 3.64%), respectively, while Pb_2Cd_2 gave the lowest content of potassium element of 2.66%. As for the results of the bi-interaction between lead element and inoculation treatment with Mycorrhiza, M_1Pb_0 treatment was excelled on the other treatments by giving it the highest potassium content of 3.93% compared to the M_0Pb_2 treatment, which gave the lowest percentage of potassium, amounted to 2.17%. The bi-

Table 1 : Effect of lead, Cadmium, inoculation with Mycorrhiza and their interaction in shoot group content of the percentage of K, P, N% elements in Canna plant.

K%				P%				N%				Treatment	
Cd*M	Pb ₂	Pb ₁	Pb ₀	Cd*M	Pb ₂	Pb ₁	Pb ₀	Cd*M	Pb ₂	Pb ₁	Pb ₀		
2.75	2.41	2.78	3.05	0.275	0.270	0.256	0.300	1.79	1.72	1.63	2.01	Cd ₀	M ₀
2.38	2.08	2.20	2.85	0.307	0.296	0.320	0.306	1.56	1.31	1.49	1.88	Cd ₁	
2.16	2.01	2.11	2.37	0.260	0.223	0.286	0.270	1.31	1.24	1.41	1.29	Cd ₂	
4.26	3.86	4.51	4.40	0.358	0.336	0.370	0.370	2.07	1.91	1.92	2.36	Cd ₀	M ₁
3.68	3.45	3.51	4.09	0.345	0.316	0.366	0.353	1.86	1.43	1.67	2.47	Cd ₁	
3.36	3.32	3.44	3.32	0.303	0.273	0.303	0.333	1.39	1.31	1.50	1.37	Cd ₂	
0.08	0.15			0.014	0.024			0.06	0.11			L.S.D 0.05	
3.50	3.14	3.64	3.72	0.317	0.303	0.313	0.335	1.93	1.82	1.77	2.19	Cd ₀	
3.03	2.77	2.85	3.47	0.326	0.306	0.343	0.330	1.71	1.37	1.58	2.18	Cd ₁	
2.76	2.66	2.78	2.85	0.281	0.248	0.295	0.301	1.35	1.27	1.46	1.33	Cd ₂	
0.06	0.10			0.010	0.017			0.04	0.08			L.S.D 0.05	
2.43	2.17	2.36	2.76	0.281	0.263	0.287	0.292	1.55	1.42	1.51	1.73	M ₀	
3.77	3.54	3.82	3.93	0.335	0.308	0.346	0.352	1.77	1.55	1.70	2.07	M ₁	
0.05	0.08			0.008	0.014			0.03	0.06			L.S.D 0.05	
	2.85	3.09	3.34		0.286	0.317	0.322		1.49	1.60	1.90	Pb	
		0.06				0.010				0.04		L.S.D 0.05	

interaction between cadmium and mycorrhiza showed to significantly excelling M_1Cd_0 treatment on the rest of the treatments, which recording 4.26%, while M_0Cd_0 gave the lowest percentage of potassium 2.16%. The results of the triple interaction between lead, Cadmium and Mycorrhiza showed that $M_1Pb_1Cd_0$ and $M_1Pb_0Cd_0$ treatments were significantly excelled by giving them the highest percentage of potassium (4.51, 4.40%) respectively. While the percentage of potassium was reduced to reach 2.01% at $M_0Pb_2Cd_2$ treatment.

Lead element concentration in shoot group (mg.Kg⁻¹ dry weight)

Table 2 shows the concentration of lead in shoot group of the plant. The results showed significant differences between the concentrations of lead element in the shoot group, the Pb_2 treatment was significantly excelled by giving it the highest concentration of lead in the shoot group of 64.66 mg.Kg⁻¹dry weight. The concentrations of the lead element in the shoot group increased by increasing concentrations of the lead elements added to the soil. This indicates the ability of canna plant to accumulate the heavy elements. The accumulated plants contain the mechanisms of resistance to these elements. So it's Phytochelatins and formation complexities with the protein known as Metallothionines. So that the element

becomes inert within the vacuoles (Kramer *et al.*, 2000; Subhashini and Swamy, 2014). The vacuoles in plants are the main store of organic acids and there is a correlation between the heavy elements and organic acids and confirms that the removal of the toxicity of such elements is because of the retention in the vacuoles and there are several strategies for plants in the accumulation of heavy elements, for example, away from the protoplasm (apoplast) or linked to the cell wall. As for the effect of Cadmium levels on lead element concentrations, there are no significant differences between them. In terms of the Mycorrhiza factor, the non-inoculated treatment M_0 was significantly excelled by giving it the highest concentration of lead in the shoot group for plant (51.77 mg.Kg⁻¹ dry weight) compared with plant inoculated with Mycorrhiza fungi, which gave the lowest concentration of lead in the shoot group was 11.67 mg.Kg⁻¹ dry weight. It is due to the fact that Mycorrhiza fungi play an important role in the removal of heavy metals and prevent the transport of the lead element to the shoot group. One of its mechanisms is to bind the lead element with the glycoprotein (glomalin), produced by the hypha of external fungus, thus reducing the presence of these elements in the soil (Bano and Darima, 2013). The results of the bi-interaction between the lead and Cadmium led to the superiority of Pb_2Cd_2 ,

Table 2 : Effect of lead, Cadmium, inoculation with Mycorrhiza and their interaction in the content of shoot and root group of canna plant from the lead element.

Lead in root group (mg.kg ⁻¹)				Lead in shoot group (mg.kg ⁻¹)				Treatment	
Cd*M	Pb ₂	Pb ₁	Pb ₀	Cd*M	Pb ₂	Pb ₁	Pb ₀		
15.80	31.75	14.71	0.88	49.46	106.25	41.03	1.10	Cd ₀	M ₀
13.48	32.01	8.14	0.30	52.84	106.25	50.33	1.94	Cd ₁	
16.02	32.11	15.68	0.26	53.01	107.50	50.07	1.46	Cd ₂	
53.03	118.75	53.37	1.95	11.28	20.34	13.50	0.00	Cd ₀	M ₁
56.66	117.75	50.62	1.60	12.33	21.95	15.03	0.00	Cd ₁	
60.02	116.83	61.37	1.84	11.39	25.66	8.52	0.00	Cd ₂	
3.64		6.30		3.42		5.93		D 0.05.S.L	
36.90	75.25	34.04	1.42	30.37	63.30	27.27	0.55	Cd ₀	
35.07	74.88	29.38	0.95	32.58	64.10	32.68	0.97	Cd ₁	
38.02	74.47	38.53	1.05	32.20	66.58	29.29	0.73	Cd ₂	
2.57		4.45		N.S		4.19		D 0.05.S.L	
15.09	31.95	12.84	0.48	51.77	106.67	47.14	1.50	M ₀	
58.23	117.78	55.12	1.80	11.67	22.65	12.35	0.00	M ₁	
2.10		3.64		1.97		3.42		D 0.05.S.L	
	74.87	33.98	1.14		64.66	29.75	0.75	Pb	
		2.57				2.42		D 0.05.S.L	

Table 3 : Effect of lead, Cadmium, inoculation with Mycorrhiza and their interaction in the content of shoot and root group of canna plant from the Cadmium element.

Cadmium Root group (mg.kg ⁻¹)				Cadmium Shoot group (mg.kg ⁻¹)				Treatment	
Cd*M	Pb ₂	Pb ₁	Pb ₀	Cd*M	Pb ₂	Pb ₁	Pb ₀		
0.17	0.14	0.20	0.18	0.20	0.35	0.14	0.10	Cd ₀	M ₀
4.30	4.15	4.32	4.43	3.45	3.41	3.47	3.47	Cd ₁	
8.26	8.24	8.24	8.31	6.35	6.42	5.75	6.89	Cd ₂	
0.19	0.18	0.23	0.17	0.26	0.30	0.38	0.11	Cd ₀	M ₁
5.94	6.20	4.70	6.92	2.68	2.20	3.29	2.55	Cd ₁	
13.85	13.83	14.56	13.16	4.38	4.08	4.99	4.08	Cd ₂	
0.68	1.18			0.37	0.65			L.S.D 0.05	
0.18	0.16	0.22	0.18	0.23	0.32	0.26	0.10	Cd ₀	
5.12	5.17	4.51	5.67	3.06	2.80	3.38	3.01	Cd ₁	
11.06	11.03	11.40	10.73	5.37	5.25	5.37	5.48	Cd ₂	
0.48	0.83			0.26	0.46			L.S.D 0.05	
4.24	4.18	4.25	4.30	2.44	3.39	3.12	3.49	M ₀	
6.66	6.73	6.50	6.75	3.33	2.19	2.89	2.24	M ₁	
0.39	0.68			0.21	0.37			L.S.D 0.05	
	5.45	5.37	5.53		2.79	3.00	2.86	Pb	
	N.S				N.S			L.S.D 0.05	

Pb₂Cd₁ and Pb₂Cd₀ treatments significantly by recording it the highest percentage of lead element in the shoot group (66.58, 64.10, 63.30 mg.Kg⁻¹ dry weight). The bi-interaction between lead concentrations and Mycorrhiza led to significantly excelling M₀Pb₂ treatment by giving it the highest concentration of lead element in the shoot group was 106.66 mg.Kg⁻¹ dry weight. The effect of the bi-interaction between Cadmium concentrations and Mycorrhiza in lead element concentrations, M₀Cd₂ and M₀Cd₁ treatment were significantly excelled by giving them the highest concentration of lead in the shoot group (53.01, 52.84 mg.Kg⁻¹ dry weight), respectively. For the three study factors, M₀Pb₂Cd₂, M₀Pb₂Cd₁ and M₀Pb₂Cd₀ were significantly excelled by recording them the highest concentration of lead in the shoot group was (107.50, 106.25 and 106.25 mg.Kg⁻¹ dry weight), while the M₁Pb₀Cd₀ treatment gave the lowest level of lead in the shoot group was (0 mg.Kg⁻¹ dry weight).

Concentration of lead element in the root group (mg.Kg⁻¹ dry weight)

Table 2 that treatment with different concentration of lead (Pb₁ and Pb₂) led to the accumulation of this element in the root group of canna plant. The 150 mg.kg⁻¹ concentration was significantly excelled in the Pb₂ treatment that recorded 74.87 mg.Kg⁻¹ dry weight and

(33.98 mg.Kg⁻¹dry weight) in Pb₂ treatment. This may be because plant may be return to the Phytoextraction group, which absorbs the heavy elements found in the soil and collects them in plant tissues. Several studies have also confirmed that the accumulate of the lead element in the roots is based on the binding of exchangeable lead ions on cell walls and sedimentation in the form of lead carbonates in cell walls (Dushenkov *et al.*, 1995). As for the effect of Cadmium concentrations, Cd₂ and Cd₀ treatments were significantly excelled by giving them the highest values in concentration of lead element in the root group (38.02, 36.90 mg.Kg⁻¹dry weight). It was also observed that the treatment of inoculation with Mycorrhiza affected the concentration of lead. The treatment M₁ was significantly excelled with the highest concentration of lead element in the root group of 58.23 mg.Kg⁻¹dry weight compared to M₀ treatment which gave (15.09 mg.Kg⁻¹dry weight). The accumulation of lead in the root group may be due to inoculation with Mycorrhiza, as it binds the lead element to the glycoprotein (glomalin), produced by the external hypha, thus reducing the amount of lead in the soil (Bano and Darima, 2013). The bi-interaction between lead element and Mycorrhiza led to significant excelling for Pb₂Cd₀, Pb₂Cd₁ and Pb₂Cd₂ treatment which recorded (75.25, 74.88, 74.47 mg.Kg⁻¹dry weight), respectively. As well

as the bi-interaction between lead concentrations and Mycorrhiza, M_1Pb_2 treatment was significantly excelled by recording it the highest concentration of lead in the root group of 117.78 mg.Kg⁻¹ dry weight. The bi-interaction between Cadmium and Mycorrhiza showed that M_1Cd_2 and M_1Cd_1 were significantly excelled by giving them the highest concentration of lead in the root group of (60.02, 56.66 mg.Kg⁻¹ dry weight), respectively. With respect to the triple interaction between lead, Cadmium and Mycorrhiza factors, $M_1Pb_2Cd_0$, $M_1Pb_2Cd_1$ and $M_1Pb_2Cd_2$ treatments were significantly excelled by giving them the highest concentration of lead in the root group of (118.75, 117.75, 116.83 mg.Kg⁻¹ dry weight), respectively.

Concentration of Cadmium element in shoot group (mg.kg⁻¹)

Table 3 shows the results of the statistical analysis of the Cadmium concentration in the shoot group of the plant. There was no significant difference between the lead concentrations added to the soil in the Cadmium element. As for Cadmium, the Cd_2 treatment was significantly excelled by giving it the highest concentration of Cadmium in the shoot group (5.37 mg.kg⁻¹). The results of this study agree with Al-Hassoun, 2015) that the increase of Cadmium concentrations in the soil cultivated with wheat led to increased its concentrations in the grain, this may be due to the tolerance of canna plant to Cadmium and the increase of the activities of enzymes antioxidant, so one of the mechanisms of tolerance of plants to toxic elements is the increased levels of antioxidant enzymes including the enzyme pyroxidase and polyphenols oxidase (Ferrol and Paola, 2016). In terms of treatment of Mycorrhiza, the M_1 treatment was significantly excelled by giving it the highest concentration of Cadmium in the shoot group (3.33 mg.Kg⁻¹ dry weight) compared to non-inoculated treatment (2.44 mg.Kg⁻¹ dry weight). The inoculation plants with Mycorrhiza led to increased absorption of heavy metals by the plant, this result agree with Kormanik *et al.*, 1980), where external mycelia was found to be responsible for transferring Cadmium from soil to plant. The results of the bi-interaction between lead and Cadmium showed that Pb_0Cd_2 , Pb_1Cd_2 and Pb_2Cd_2 treatments were significantly excelled by giving them the highest concentration of Cadmium in the shoot group (5.48, 5.37, 5.25 mg.kg⁻¹). In terms of the bi-interaction between the concentration of the lead element and the inoculation with the Mycorrhiza, the M_0Pb_0 treatment was significantly excelled with the value of (3.49 mg.kg⁻¹ dry weight). In the bi-interaction between Cadmium concentrations and Mycorrhiza, M_0Cd_2 treatment was significantly excelled

by giving it the highest concentration of Cadmium in the shoot total of (6.35 mg.kg⁻¹ dry weight). The results of the triple interaction between the factors of the study showed that the M_0Pb_0Cd and $M_0Pb_2Cd_2$ treatments was excelled by giving them the highest concentrations of Cadmium in the shoot group (6.89, 6.42 mg.kg⁻¹).

Concentration of Cadmium element in the root group (mg.kg⁻¹)

Table 3 shows concentrations of Cadmium in the root group of the *Canna generalis* plant, lead treatments were not significantly different in the content of the root group of Cadmium. As for Cadmium treatments, Cd_2 treatment was significantly excelled by giving it the highest concentration of Cadmium in the root group of (11.06 mg.kg⁻¹ dry weight). The results of this study are consistent with Al-Khuzai, 2012). In his study on the plant of the sun flower, as it was found that this plant has good ability to collect the Cadmium and its accumulation in its different tissues and the root group excelled on the shoot group in accumulation and assembly, also this results agrees with Wiessmann and Nehring, 1960) showed that the highest concentration of Cadmium was in the root of indica canna plant and the concentration of the element in the soil was reduced after the end of the experiment and this is evidence of the absorption of Cadmium by the plant. The effect of plant inoculation by Mycorrhiza, The M_1 treatment was significantly excelled with the highest concentration of Cadmium in the root group of 6.66 mg.kg⁻¹ dry weight compared with M_0 treatment which recorded 4.23 mg.kg⁻¹ dry weight. The increase in Cadmium concentrations when plant inoculation by Mycorrhiza may be due to the association of Cadmium with the glycoprotein (glomalin), which is produced by the external hypha, thereby reducing the amount of Cadmium in the soil and thereby increasing its presence in the roots (Bano and Darima, 2013). In terms of the bi-interaction between lead and Cadmium, Pb_1Cd_2 , Pb_2Cd_2 and Pb_0Cd_2 treatments were significantly excelled by giving them the highest concentration of Cadmium in the root group of (11.40, 11.03, 10.73 mg.kg⁻¹ dry weight), respectively. The effect of the bi-interaction between lead concentration and Mycorrhiza at Cadmium levels in the root group, M_1Pb_0 and M_1Pb_2 were significantly excelled by giving them the highest concentration of Cadmium in the root group of (6.75, 6.73 mg.kg⁻¹). The results of the bi-interaction between Cadmium and Mycorrhiza concentrations showed significant excelling of the M_1Cd_2 treatment, which recorded the highest concentration of Cadmium in the root group of 13.85 mg.kg⁻¹. In the triple interaction between the study factors, $M_1Pb_1Cd_2$ and $M_1Pb_2Cd_2$ were significantly excelled by giving them the

Table 4 : Concentrations of lead (Pb) and Cadmium (Cd) availability in the soils cultivated with canna plants after the end of experiment (mg.kg⁻¹).

Cadmium availability after the end of experiment (mg.kg ⁻¹)				Lead availability after the end of experiment (mg.kg ⁻¹)				Treatment	
Cd*M	Pb ₂	Pb ₁	Pb ₀	Cd*M	Pb ₂	Pb ₁	Pb ₀		
0.123	0.117	0.150	0.103	0.467	0.450	0.500	0.450	Cd ₀	M ₀
0.183	0.117	0.283	0.150	0.458	1.023	0.233	0.117	Cd ₁	
0.339	0.250	0.367	0.400	0.662	1.373	0.467	0.147	Cd ₂	
0.173	0.183	0.200	0.137	0.550	0.400	1.100	0.150	Cd ₀	M ₁
0.461	0.500	0.350	0.533	0.706	1.117	0.700	0.300	Cd ₁	
0.567	0.300	0.700	0.700	0.641	1.200	0.550	0.173	Cd ₂	
0.133	0.231			0.209	0.362			L.S.D0.05	
0.148	0.150	0.175	0.120	0.508	0.425	0.800	0.300	Cd ₀	
0.322	0.308	0.317	0.341	0.582	1.070	0.467	0.208	Cd ₁	
0.453	0.275	0.533	0.550	0.652	1.287	0.508	0.160	Cd ₂	
0.094	0.212			N.S	0.256			L.S.D0.05	
0.215	0.161	0.267	0.218	0.529	0.949	0.400	0.238	M ₀	
0.400	0.328	0.417	0.457	0.632	0.906	0.783	0.208	M ₁	
0.077	0.133			0.120	0.209			L.S.D0.05	
	0.244	0.341	0.337		0.927	0.592	0.223	Pb	
	0.094				0.147			L.S.D0.05	

highest concentration of Cadmium in the root group of (14.56, 13.83 mg.kg⁻¹ dry weight), respectively.

Lead element preparing in soil after experiment (mg.kg⁻¹)

Table 4 shows the concentrations of the available lead element in the soil cultivated with the Canna plant after the end of the experiment. The results indicate that there are significant differences between the concentrations of the available lead element in the soil with the concentration of the available lead element in the treatment Pb₂ (0.927 mg.kg⁻¹ Soil). That the increasing of lead in the soil may be due to increased concentrations of lead that was added to soil before cultivating. These results are consistent, who explained that increasing concentrations of lead in the soil increases the availability and according to levels of addition or may be due to the role of peat moss (organic matter) in reducing soil pH and then increase the availability of this element, noting the concentrations values of Lead element in soils cultivated with canna plants did not exceed the critical limit of (WHO/ FAO, 2007) were (100 mg.kg⁻¹) of lead in soil after the end of the experiment. As for the effect of Cadmium concentrations on the lead element, there is no significant difference between them. The effect of

plant inoculation with Mycorrhiza increased lead element availability, the soil inoculated with Mycorrhiza showed an increased in the lead element availability in it which gave 0.632 mg.kg⁻¹ soil compared with the non-inoculated plants M₀, which recorded 0.529 mg.kg⁻¹ soil. It is probably due to the role of Mycorrhiza fungus in the reduction pH of the soil and thus increasing element availability. As for the effect of the bi-interaction between lead and Cadmium elements, Pb₂Cd₂ and Pb₂Cd₁ treatments are significantly excelled, which did not differ significantly between them, by giving them the highest concentration of lead (1.287, 1.070 mg.kg⁻¹ soil), respectively. In the bi-interaction between lead concentrations and Mycorrhiza, M₀Pb₂, M₁Pb₂ and M₁Pb₁ treatments were significantly excelled which recorded values (0.949, 0.906, 0.783 mg.kg⁻¹ soil), respectively. In the bi-interaction between Cadmium and Mycorrhiza, the M₁Cd₁ treatment is significantly excelled by giving it the highest concentration of lead in the soil (0.706 mg.kg⁻¹ soil) which was not significantly different from M₀Cd₂, M₁Cd₂ and M₁Cd₀, that recorded values (0.662, 0.641, 0.550 mg.kg⁻¹ soil). The triple interaction between the study factors showed that M₀Pb₂Cd₂, M₁Pb₂Cd₂, M₁Pb₂Cd₁ and M₁Pb₂Cd₀ were significantly excelled in increasing the concentrations of lead element

availability which recorded the values (1.373, 1.200, 1.117, 1.100 mg.kg⁻¹ soil), respectively.

Cadmium element in the soil before end of experiment (mg.kg⁻¹)

Table 4 indicates significant differences in concentrations of Cadmium element availability in the soil. It was observed that the levels of the lead element had a significant effect on Cadmium availability in the soil. The Pb₁ and Pb₀ treatments were excelled by increasing the Cadmium element availability in soil which recorded (0.341, 0.337 mg.kg⁻¹ soil). As for the effect of Cadmium concentrations on the Cadmium element availability in soil, the Cd₂ treatment was significantly excelled by giving it the highest concentration of the Cadmium element availability (0.453 mg.kg⁻¹ soil). The increase in the Cadmium element availability may be due to the increased concentrations added to soil, This agree with Abd Al-Latif and Ali, (2016) or possibly due to the role of potassium sulphate fertilizer which reduced soil pH and thus increased the availability of this element in soil, when comparing these values with those of (WHO / FAO, 2007), they did not exceed the permissible limit of 3 mg.kg⁻¹ of Cadmium in the soil after the end of the experiment. Plant inoculation with Mycorrhiza showed increasing Cadmium element availability. The M₁ treatment is significantly excelled by giving it the highest concentration of Cadmium availability (0.400 mg.kg⁻¹ soil) compared to M₀, which recorded (0.215 mg.kg⁻¹ soil). This is due to the role of Mycorrhiza fungi in reducing soil pH and thus increasing its availability. Pb₀Cd₂ and Pb₁Cd₂ treatments which did not differ significantly between them recording (0.550, 0.533 mg.kg⁻¹ soil) in the bi-interaction between lead and Cadmium. The bi-interaction between lead concentrations and Mycorrhiza, M₁Pb₀ and M₁Pb₁ treatments was significantly excelled, which recorded the highest concentration of Cadmium element availability (0.457, 0.417 mg.kg⁻¹ soil) and which did not significantly differ from the M₁Pb₂ treatment (0.328 mg.kg⁻¹ soil). As for the bi-interaction between the Cadmium concentrations and Mycorrhiza, M₁Cd₂ and M₁Cd₁ treatments excelled, which did not differ significantly between them in recording the highest concentration of Cadmium element availability (0.567 and 0.461 mg.kg⁻¹ soil). The triple interaction between the factors of the study showed the significant superiority of M₁Pb₀Cd₂, M₁Pb₁Cd₂ and M₁Pb₀Cd₁ treatments by giving them the highest concentrations of Cadmium availability in the soil (0.700, 0.700, 0.533 mg.kg⁻¹ soil).

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